REMARKS

Claims 1-10, 14, 16 and new Claims 58-59 remain pending in the instant application after the foregoing amendments. Claims 15, 19-23, 39-40 have been cancelled. Applicants expressly reserve the right to file continuation applications directed to the subject matter not currently being pursued.

Specification

The Examiner has objected to the specification because pages allegedly contained illegible text. While Applicants contend that the specification submitted was legible and accurate, copies of the pages cited by the Examiner have been attached as requested.

Section 112, First Paragraph

The Examiner has rejected Claims 15-16, 19-23 and 39-40 under 35 U.S.C. §112, first paragraph, as allegedly not being enabling for the methods of treatment.

In order to expedite the prosecution of this application, Applicants note that Claims 15, 19-23 and 39-40 have been canceled. Applicants expressly reserve the right to file continuation applications directed to the subject matter not currently being pursued. Applicants have amended Claim 16 and respectfully assert that the instant application is enabling for the method of treating the specific cancers listed. Additionally, Applicants have added new Claims 58 and 59. Applicants respectfully assert that no new matter was added by this amendment. As noted by the Examiner, there is support in the application for the method of treating lung adenocarcinoma, which is new Claim 58. Additionally, in support of a method of treating acute myeloid leukemia, applicants have disclosed assays using two VEGF kinase receptors, Flt-1 and Flt-3. (See specification p. 47-49). It is known in the art that VEGF and Flt-1/Flt-3 receptors are associated with acute myeloid leukemia. Hu et al., *Solubale Vascular Endothelial Growth Factor Receptor 1 and Not Receptor 2, Is an Independent Prognostic Factor in Acute Myeloid Leukemia and Myelodysplastic Syndromes*, Cancer, 100(9):1884-91 (2004). Hu et al reports that Flt-1 and not Flt-2 is involved in acute myeloid leukemia. Also regarding Flt-3, Meshinchi et al., *Activating Mutations of RTK/ras Signal Transduction Pathway in Pediatric Acute Myeloid Leukemia*, Blood, 102(4):1474-1479 (2003), reports that "Mutations in the Flt-3 receptor gene

are the most common genetic alteration in [acute myeloid leukemia], and these mutations are associated with rapid disease progression and resistance to conventional therapy." See page 1474, Introduction. Schuch et al., *In vivo Administration of Vascular Ednothelial Growth Factor (VEGF) and its antagonist, soluble neuropilin-1, predicts a role of VEGF in the Progression of Acute Myeloid Leukemia in vivo*, Blood, 100(13):4622-4628 (2002) provides in vivo data supporting VEGFr's role in acute myeloid leukemia. See generally. Based on the description in the specification supported by the state of the art, applicants have enabled the method of treating acute myeloid leukemia.

Therefore, Applicants respectfully request that the rejection based on 35 U.S.C. §112, first paragraph, be withdrawn.

Section 102

The Examiner has rejected Claims 1-5, 14-16, 19-23 and 39-40 under 35 U.S.C. 102(b) as allegedly being anticipated by Bilodeau et al., WO 02/45652 and by Das et al., WO 00/62778.

Applicants respectfully traverse this rejection. For a single reference to anticipate an invention, the reference must, either expressly or inherently, disclose each and every element of the claimed invention. *RCA Corp. v. Applied Digital Data Sys., Inc.*, 221 U.S.P.Q. 385, 388 (Fed. Cir. 1984). Applicants point out that Claims 1-5, as amended, define substituent X as O or S. Additionally, the amended claims do not allow for an amide or amine in the R^1 or R^2 position of Formula I. Unlike the claims of the instant application, the compounds disclosed in the references cited by the Examiner require two amine groups, one linking the thiazole and pyrimidine and one amine linking the pyrimidine to other substituents (which would be equivalent to the R^5 position of claimed Formula I). The claims of the instant application require either an oxygen or sulfur atom in that position. Additionally, the compounds disclosed in Das et al. in Examples 44-472 require an amide in the R^2 position of Formula I of the instant application, whereas the claimed invention does not allow for that substitution. Because each and every element is not disclosed in the cited references, Applicants assert that the references do not anticipate the claimed invention. Therefore, Applicants respectfully request that this rejection be withdrawn.

Section 103

The Examiner has rejected Claims 1-4, 14-16, 19-23 and 39-40 under 35 U.S.C. 103(a) as allegedly being unpatentable over Das et al.

Applicants respectfully traverse this rejection. As noted above, Claims 1-4 define substituent X as either O or S, and not N. These claims also do not allow for a substituted amide group in the R^2 position of Formula I. Das et al. does not teach, nor suggest, replacing the N at the "X" position with an oxygen or sulfur. Formula I of Das et al. requires an amine group on the right side of substituent Q. It does not allow for a non-amine moiety in this position, nor does it teach or suggest such a modification. Because one with ordinary skill in the art would not be motivated by Das et al. to modify the compounds disclosed therein to arrive at the claimed invention, the reference does not render the instant invention obvious.

Additionally, the Examiner has rejected Claims 1-10, 14-16, 19-23 and 39-40 under 35 U.S.C. 103(a) as allegedly being unpatentable over Bilodeau et al.

Applicants respectfully traverse this rejection. As noted above, Claims 1-5 define substituent X as either O or S, and not N. Additionally, Claim 6 has been amended to claim compounds that possess an oxygen or sulfur atom in the "X" position. Compounds 19-3 and 19-4 of Bilodeau et al. does not teach, nor suggest, replacing the N at the "X" position with an oxygen or sulfur atom. One with ordinary skill in the art would not be motivated by Bilodeau et al. to modify the compounds disclosed therein to arrive at the claimed invention

Applicants believe that the Examiner's concerns have been addressed and respectfully request that the rejections under 35 U.S.C. 103(a) be withdrawn.

If a telephonic communication with the Applicants' representative will advance the prosecution of the instant application, please telephone the representative indicated below. Applicants believe no additional fees are due but the Commissioner is authorized to charge any fees required in connection with this amendment to Merck Deposit Account No. 13-2755.

Respectfully submitted,

By Dianne Brown

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Attorney for Applicant(s)

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Date: July 20, 2006

Attachment

cancers. Kamb, A. et al. (1994) *Science* 264, 436-440, Nobori, et al. (1994) *Nature* 368, 753-756, Spruck, C.H. et al. *Nature* 370, 183-184, Hunter, T. and Pines, J. (1991) *Cell* 66, 1071-1074, Keyomarsi, K. and Pardee, A.B. (1993) *Proc. Natl. Acad. Sci. U.S.A.* 90, 1112-1116 and Wang, T.C. (1994) *Nature* 369, 669-671.

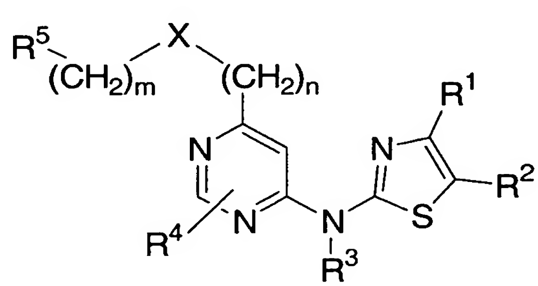
Members of the cyclin dependent kinase family include Cdk2 and Cdk4. Both are active in the G_1 phase of cell cycle and regulate entry into the G_1/S phase transition. In one pathway, these kinases regulate the phosphorylation of the retinoblastoma protein. Substrate phosphorylation releases the E2F transcription factor which in turn regulates the expression of genes required for S phase entry. Inhibition of these kinases, therefore, blocks cell entry into the S phase and downstream proliferative events.

Small molecular cyclin dependent kinase inhibitors have already been identified and shown to have growth inhibitory activity against a number of different tumor types *in vitro* and *in vivo*. Glab, N. et al. (1994) *FEBS Lett.* 353, 207-211, Kitagawa, M. et al. (1993) *Oncogene* 8, 2425-2432, Losiewicz, M.D. et al. (1994) *Biochem. Biophys. Res. Commun.* 201, 589-595, Carlson, B.A. et al. (1996) *Cancer Res.* 56, 2973-2978, Kelland, L.R. (2000) *Expert Opin. Invest. Drugs* 9, 2903-2911 and Senderowicz, A.M. (1999) *Invest. New Drugs* 17, 313-320.

Accordingly, the identification of small compounds which specifically inhibit, regulate and/or modulate the signal transduction of kinases is desirable and is an object of this invention.

SUMMARY OF THE INVENTION

The present invention relates to compounds that are capable of inhibiting, modulating and/or regulating signal transduction of kinases. One embodiment of the present invention is illustrated by a compound of Formula I, and the pharmaceutically acceptable salts and stereoisomers thereof:



I

2-[(6-{[(1,1-dioxidotetrahyrothien-3ylmethyl]amino}-2-methylpyrimidin-4-yl) amino]-1,3-thiazole-5-carbonitrile;
2-({6-[(1,4-dioxan-2-ylmethyl)amino]-2-methylpyrimidin-4-yl}amino)-1,3-thiazole-5-carbonitrile;
2-({6-[(3-morpholin-4-ylpropyl)amino]pyrimidin-4-yl}amino)-1,3-thiazole-5-carbonitrile;
2-[-({6-[5-cyano-1,3-thiazol-2-ylamino]-2-methylpyrimidin-4-yl}amino)piperidin-1-yl]-N-isopropylacetamide;
tert-butyl-4-({6-[(5-cyano-1,3-thiazol-2-ylamino]-2-methylpyrimidin-4-yl}amino) piperidine-1-carboxylate;
2-{[2-methyl-6-(piperidin-4-ylamino)pyrimidin-4-yl]amino}-1,3-thiazole-5-carbonitrile;
tert-butyl-4-({6-[(5-cyano-1,3-thiazol-2-yl)amino]methyl}-2-methylpyrimidin-4-yl} amino) piperidine-1-carboxylate;
2-({2-methyl-6-[(piperidin-4-ylmethyl)amino]pyrimidin-4-yl}amino)-1,3-thiazole-5-carbonitrile;
2-{[5-methyl-6-(piperidin-4-ylamino)pyrimidin-4-yl]oxy}-1,3-thiazole-5-carbonitrile;
tert-butyl-2-[({6-[(5-cyano-1,3-thiazol-2-yl)amino]-2-methylpyrimidin-4-yl}oxy) methyl]-morpholine-4-carboxylate;
2-{[2-methyl-6-(morpholin-2-ylmethoxy)pyrimidin-4-yl]amino}-1,3-thiazole-5-carbonitrile;
2-{[2-methyl-6-(tetrahydro-2-pyran-4-yloxy)pyrimidin-4-yl]amino}-1,3-thiazole-5-carbonitrile;
2-{[2-isopropyl-6-(piperidin-4-yloxy)pyrimidin-4-yl]amino}-1,3-thiazole-5-carbonitrile;
2-({6-[(1,1-dioxidotetrahydrothien-3-yl)amino]-2-methylpyrimidin-4-yl}amino)-1,3-thiazole-5-carbonitrile;
2-{[2-methyl-6-(tetrahydrofuran-3-ylamino)pyrimidin-4-yl]amino}-1,3-thiazole-5-carbonitrile;
tert-butyl{4-[({6-[(5-cyano-1,3-thiazol-2-yl)amino]-2-methylpyrimidin-4-yl}oxy) methyl]piperidin-1-yl}acetate;
{4-[({6-[(5-cyano-1,3-thiazol-2-yl)amino]-2-methylpyrimidin-4-yl}oxy)methyl] piperidin-1-yl}acetic acid;
N-(tert-butyl)-2-{4-[({6-[(5-cyano-1,3-thiazol-2-yl)amino]-2-methylpyrimidin-4-yl} oxy)methyl]piperidin-1-yl}acetamide;
2-({2-methyl-6-[(2-morpholin-4-ylethyl)thio]pyrimidin-4-yl}amino)-1,3-thiazole-5-carbonitrile;
and
2-{[6-(piperidin-4-ylthio)pyrimidin-4-yl]amino}-1,3-thiazole-5-carbonitrile;
or a pharmaceutically acceptable salt or stereoisomer thereof.

Yet another embodiment of the present invention is a compound which is: 2-({2-methyl-6-[(3S)-pyrrolidin-3-yloxy]pyrimidin-4-yl}amino)-1,3-thiazole-5-carbonitrile

compounds can inhibit VEGF-induced vascular permeability and therefore serve to prevent or treat blood-brain barrier disruption associated with bacterial meningitis.

The present invention further encompasses a method to treat or prevent endometriosis comprised of administering a therapeutically effective amount of a claimed compound. An increase in VEGF expression and angiogenesis is associated with the progression of endometriosis (Stephen K. Smith, *Trends in Endocrinology & Metabolism*, Vol. 12, No. 4, May/June 2001). Inhibition of VEGF by the current compounds would therefore inhibit angiogenesis and treat endometriosis.

A further embodiment of the present invention is a method of treating acute myeloid leukemia (AML) which comprises administering a therapeutically effective amount of a claimed compound. Activation of FLT3 on leukemic cells by FLT3 ligand leads to receptor dimerization and signal transduction in pathways that promote cell growth and inhibit apoptosis (*Blood,* Vol. 98, No. 3, pp.885-887 (2001)). The present compounds are therefore useful to treat AML via inhibition of the tyrosine kinase domain of Flt-3.

Another embodiment of the present invention is a method of treating or preventing cancer via the dual inhibition of cyclin dependent kinase and tyrosine kinase. Cyclin dependent kinases are known to regulate cell cycle progression and cyclin dependent kinase inhibitors have been shown to block cell proliferation. As described above, inhibition of tyrosine kinases is useful in the treatment and prevention of cancer. Furthermore, inhibition of cyclin dependant kinases is also useful in the treatment and prevention of cancer (Glab et al., *FEBS Lett.* 353, 207-211 (1994), Kitagawa et al., *Oncogene.* 8, 2425-2432 (1993), Losiewicz et al., *Biochem. Biophys. Res. Commun.* 201, 589-595 (1994), Carlson et al. *Cancer Res.* 56, 2973-2978 (1996), Kelland, L.R. *Expert Opin. Invest. Drugs.* 9, 2903-2911 (2000) and Senderowicz, A.M. *Invest. New Drugs.* 17, 313-320 (1999)). Thus, dual inhibition of two separate signaling pathways provides an advantage in the inhibition of cell proliferation and cancer progression. The present compounds demonstrate dual inhibitory activity against cyclin dependent kinases as well as tyrosine kinases and are therefore useful in the treatment and prevention of cancer.

The present invention encompasses a method of treating or preventing cancer via the dual inhibition of tyrosine kinase and cyclin dependent kinase wherein the dual inhibitor is selected from:

tert-butyl{4-[({6-[(5-cyano-1,3-thiazol-2-yl)amino]-2-methylpyrimidin-4-yl}oxy) methyl]piperidin-1-yl}acetate;

2-{[2-methyl-6-(piperidin-4-yloxy)pyrimidin-4-yl]amino}-1,3-thiazole-5-carbonitrile;

2-{[2-methyl-6-(piperidin-4-ylmethoxy)pyrimidin-4-yl]amino}-1,3-thiazole-5-carbonitrile; and

2-{[2-isopropyl-6-(piperidin-4-yloxy)pyrimidin-4-yl]amino}-1,3-thiazole-5-carbonitrile;

chloropyridin-2-ylmethyl)-piperidine-1-ylmethyl]-2-methylimidazol-1-ylmethyl} benzonitrile, 4-{5-[4-hydroxymethyl-4-(3-chlorobenzyl)-piperidine-1-ylmethyl]-2-methylimidazol-1-ylmethyl}benzonitrile, 4-{3-[4-(2-oxo-2H-pyridin-1-yl)benzyl]-3H-imidazol-4-ylmethyl}benzonitrile, 4-{3-[4-(5-chloro-2-oxo-2H-[1,2']bipyridin-5'-ylmethyl]-3H-imidazol-4-ylmethyl}benzonitrile, 4-{3-[4-(2-oxo-2H-[1,2'] bipyridin-5'-ylmethyl]-3H-imidazol-4-ylmethyl}benzonitrile, 4-[3-(2-oxo-1-phenyl-1,2-dihydropyridin-4-ylmethyl)-3H-imidazol-4-ylmethyl}benzonitrile, 18,19-dihydro-19-oxo-5*H*,17*H*-6,10:12,16-dimetheno-1H-imidazo[4,3-*c*][1,11,4]dioxaazacyclo-nonadecine-9-carbonitrile, (±)-19,20-dihydro-19-oxo-5*H*-18,21-ethano-12,14-etheno-6,10-metheno-22*H*-benzo[*d*]imidazo[4,3-*k*][1,6,9,12]oxatriaza-cyclooctadecine-9-carbonitrile, 19,20-dihydro-19-oxo-5*H*,17*H*-18,21-ethano-6,10:12,16-dimetheno-22*H*-imidazo[3,4-*h*][1,8,11,14]oxatriazacycloeicosine-9-carbonitrile, and (±)-19,20-dihydro-3-methyl-19-oxo-5*H*-18,21-ethano-12,14-etheno-6,10-metheno-22*H*-benzo [*d*]imidazo[4,3-*k*][1,6,9,12]oxa-triazacyclooctadecine-9-carbonitrile.

Other examples of prenyl-protein transferase inhibitors can be found in the following publications and patents: WO 96/30343, WO 97/18813, WO 97/21701, WO 97/23478, WO 97/38665, WO 98/28980, WO 98/29119, WO 95/32987, U.S. Patent No. 5,420,245, U.S. Patent No. 5,523,430, U.S. Patent No. 5,532,359, U.S. Patent No. 5,510,510, U.S. Patent No. 5,589,485, U.S. Patent No. 5,602,098, European Patent Publ. 0 618 221, European Patent Publ. 0 675 112, European Patent Publ. 0 604 181, European Patent Publ. 0 696 593, WO 94/19357, WO 95/08542, WO 95/11917, WO 95/12612, WO 95/12572, WO 95/10514, U.S. Patent No. 5,661,152, WO 95/10515, WO 95/10516, WO 95/24612, WO 95/34535, WO 95/25086, WO 96/05529, WO 96/06138, WO 96/06193, WO 96/16443, WO 96/21701, WO 96/21456, WO 96/22278, WO 96/24611, WO 96/24612, WO 96/05168, WO 96/05169, WO 96/00736, U.S. Patent No. 5,571,792, WO 96/17861, WO 96/33159, WO 96/34850, WO 96/34851, WO 96/30017, WO 96/30018, WO 96/30362, WO 96/30363, WO 96/31111, WO 96/31477, WO 96/31478, WO 96/31501, WO 97/00252, WO 97/03047, WO 97/03050, WO 97/04785, WO 97/02920, WO 97/17070, WO 97/23478, WO 97/26246, WO 97/30053, WO 97/44350, WO 98/02436, and U.S. Patent No. 5,532,359. For an example of the role of a prenyl-protein transferase inhibitor on angiogenesis see *European J. of Cancer*, Vol. 35, No. 9, pp.1394-1401 (1999).

Examples of HIV protease inhibitors include amprenavir, abacavir, CGP-73547, CGP-61755, DMP-450, indinavir, nelfinavir, tipranavir, ritonavir, saquinavir, ABT-378, AG 1776, and BMS-232,632. Examples of reverse transcriptase inhibitors include delaviridine, efavirenz, GS-840, HB Y097, lamivudine, nevirapine, AZT, 3TC, ddC, and ddI.

antiinflammatory. Steroidal anti-inflammatories include, but are not limited to, corticosteroids, mineralocorticoids, dexamethasone, prednisone, prednisolone, methylpred, and betamethasone. This combination is particularly useful in ophthalmic formulations which may, in some cases, be associated with irritation of the ocular tissues.

A particularly useful combination for the treatment of diseases wherein aberrant angiogensis is present involves administering a therapeutically effective amount of the instantly disclosed kinase inhibiting compounds in combination with photodynamic therapy and a photosensitive drug such as verteoporfin (BPD-MA) (Carruth, Clinical Applications of Photodynamic Therapy, *Int. J. Clin. Pract.* 1998; 52(1):39-42). Such diseases include, but are not limited to, age-related macular degeneration (Bressler, Treatment of Age-Related Macular Degeneration with Photodynamic Therapy Investigation Using Verteoporfin, *Invest. Ophthalmol. Vis. Sci.* 1998; 39 S242), cancer, especially melanoma and non-melanoma skin cancer, including basal cell and squamous cell carcinomas, (Hassan and Parrish, Photodynamic Therpay in Cancer, *Cancer Med.* 1997; Dougherty et al., Photodynamic Therapy for the Treatment of Cancer: Current Status and Advances in Photodynamic Therapy of Neoplastic Disease. Kessel (Ed.), CRC Press, 1989; 1-19); Dougherty et al., Photodynamic Therpay, *J. Natl. Cancer Inst.*, 1998, 90(12): 889-905; Jori, Factors Controlling the Selectivity and Efficiency of Tumour Damage in Photodynamic Therapy, *Laser Med. Sci.* 1990; 5: 115-120; Zhou, Mechanism of Tumour Necrosis Induced by Photodynamic Therapy, *J. Photochem. Photobiol.* 1989; 3: 299-318), psoriasis (Bissonnette et al., Photodynamic Therapy of Psoriasis and Psoriatic Arthritis with BPD verteporfin. 7th Biennial Congress, International Photodynamic Association, Nantes, France 1998:73), and rheumatoid arthritis (Hendrich et al., Photodynamic Therapy for Rheumatoid Arthritis. *Lasermedizin* 11: 73-77 (1995); Hendrich et al. Photodynamic Laser Therapy for Rheumatoid Arthritis: Cell Culture Studies and Animal Experiments, *Knee Surg. Sports Traumatol. Arthroscopy* 5: 58-63 (1997).

Another embodiment of the instant invention is the use of the presently disclosed compounds in combination with gene therapy for the treatment of cancer. For an overview of genetic strategies to treating cancer see Hall et al (*Am J Hum Genet* 61:785-789, 1997) and Kufe et al (*Cancer Medicine*, 5th Ed, pp 876-889, BC Decker, Hamilton 2000). Gene therapy can be used to deliver any tumor suppresing gene. Examples of such genes include, but are not limited to, p53, which can be delivered via recombinant virus-mediated gene transfer (see U.S. Patent No. 6,069,134, for example), a uPA/uPAR antagonist ("Adenovirus-Mediated Delivery of a uPA/uPAR Antagonist Suppresses Angiogenesis-Dependent Tumor Growth and Dissemination in Mice," *Gene Therapy*, August 1998;5(8):1105-13), and interferon gamma (*J Immunol* 2000;164:217-222).

disclosed by Delaney et al. U.K. Patent Application No. 2,207,351 and by Haslanger et al. in U.S. Patent No. 4,749,688. Compounds possessing both neutral endopeptidase and angiotensin converting enzyme inhibition activity are disclosed by Flynn et al. in U.S. Patent No.5,366,973, European Patent Application No. 481,522 and PCT Patent Applications Nos. WO 93/16103, and WO 94/10193, Warshawsky et al. European Patent Applications Nos. 534,363, 534,396 and 534,492, Fournie- Zaluski European Patent Application No. 524,553, Karanewsky et al. European Patent Application No. 599,444, Karanewsky European Patent Application No. 595, 610, Robl et al., European Patent Application No. 629,627, Robl, U.S. Patent No. 5,362,727 and European Patent Application No. 657,453. The disclosures of all such patents and publications are incorporated herein by reference.

Further, the anti-hypertensive agents which may be used in accordance with this invention and the pharmaceutically acceptable salts thereof may occur as prodrugs, hydrates or solvates. Said hydrates and solvates are also within the scope of the present invention. Preferred anti-hypertensive agents of the invention include, calcium channel blockers, A-II antagonists, ACE inhibitors and β-blockers. More preferred anti-hypertensive agents of the invention include ACE inhibitors, particularly lisinopril, enalapril and captopril, and A-II antagonists, particularly losartan. The anti-hypertensives described herein are generally commercially available, or they may be made by standard techniques including those described in the references cited above.

The instant compounds are also useful, alone, or in combination with ovulation stimulators such as, but not limited to; bromocriptine (e.g., PARLODEL), luprolide (e.g., LUPRON), clomifene (e.g., CLOMID, SEROPHENE) and pharmaceutically acceptable salts thereof, follicle stimulating hormone (e.g., FERTINEX/ METRODIN, FOLLISTIM, GONAL F), menopausal gonadotropin or mentropins (e.g., REPRONEX), chorionic gonadotropin (e.g., PROFASI, PREGNYL), luteinizing hormone releasing hormone (e.g., GONADORELIN), luteinizing hormone and combinations thereof to treat or prevent ovarian hyper-stimulation syndrome (OHSS). OHSS is a side effect that occurs during infertility treatment with ovulation inducing drugs. OHSS has also been reported to occur as a result of increased endogenous seceretion of gonadotropins (*Obstet. Gynecol.* 21:28, 1963; *J. Obstet. Gynaecol. Br. Commonw.* 74:451, 1967). Symptoms of OHSS range from mild to critical and are associated with ovarian enlargement and increased vascular permeability. Women with the most severe symptoms demonstrate increased VEGF levels in follicular fluids that are reversed via the addition of a VEGF antibody indicating that VEGF is responsible for vascular permeabiliy contributing to the pathogenesis of OHSS. Levin, E.R. et al., *J. Clin. Invest.* 102, 1978-1985 (1998). Therefore, a method of treating or preventing ovarian hyper-stimulation syndrome, which comprises

piperidinyl, pyrrolidinyl, morpholinyl, thiomorpholinyl, dihydrobenzoimidazolyl, dihydrobenzofuranyl, dihydrobenzothiophenyl, dihydrobenzoxazolyl, dihydrofuranyl, dihydroimidazolyl, dihydroindolyl, dihydroisooxazolyl, dihydroisothiazolyl, dihydrooxadiazolyl, dihydrooxazolyl, dihydropyrazinyl, dihydropyrazolyl, dihydropyridinyl, dihydropyrimidinyl, dihydropyrrolyl, dihydroquinolinyl, dihydrotetrazolyl, dihydrothiadiazolyl, dihydrothiazolyl, dihydrothienyl, dihydrotriazolyl, dihydroazetidinyl, methylenedioxybenzoyl, tetrahydrofuranyl, and tetrahydrothienyl, and N-oxides thereof. Attachment of a heterocyclyl substituent can occur via a carbon atom or via a heteroatom.

R^5 is heterocyclyl. It is understood that attachment of any substituents may occur via a carbon atom or a heteroatom.

The alkyl, alkenyl, alkynyl, cycloalkyl, aryl, heteroaryl and heterocyclyl substituents may be substituted or unsubstituted, unless specifically defined otherwise. For example, a $(C_1\text{-}C_6)$alkyl may be substituted with one or more substituents selected from OH, oxo, halogen, alkoxy, dialkylamino, or heterocyclyl, such as morpholinyl, piperidinyl, and so on. In this case, if one substituent is oxo and the other is OH, the following are included in the definition: $-(C{=}O)CH_2CH(OH)CH_3$, $-(C{=}O)OH$, $-CH_2(OH)CH_2CH(O)$, and so on.

The pharmaceutically acceptable salts of the compounds of this invention include the conventional non-toxic salts of the compounds of this invention as formed inorganic or organic acids. For example, conventional non-toxic salts include those derived from inorganic acids such as hydrochloric, hydrobromic, sulfuric, sulfamic, phosphoric, nitric and the like, as well as salts prepared from organic acids such as acetic, propionic, succinic, glycolic, stearic, lactic, malic, tartaric, citric, ascorbic, pamoic, maleic, hydroxymaleic, phenylacetic, glutamic, benzoic, salicylic, sulfanilic, 2-acetoxy-benzoic, fumaric, toluenesulfonic, methanesulfonic, ethane disulfonic, oxalic, isethionic, trifluoroacetic and the like. The preparation of the pharmaceutically acceptable salts described above and other typical pharmaceutically acceptable salts is more fully described by Berg *et al.*, "Pharmaceutical Salts," *J. Pharm. Sci.,* 1977:66:1-19, hereby incorporated by reference. The pharmaceutically acceptable salts of the compounds of this invention can be synthesized from the compounds of this invention which contain a basic or acidic moiety by conventional chemical methods. Generally, the salts of the basic compounds are prepared either by ion exchange chromatography or by reacting the free base with stoichiometric amounts or with an excess of the desired salt-forming inorganic or organic acid in a suitable solvent or various combinations of solvents. Similarly, the salts of the acidic compounds are formed by reactions with the appropriate inorganic or organic base.

9. Add 30 μL of scintillation cocktail, seal plate and count in a Wallac Microbeta scintillation counter.

II. HUMAN UMBILICAL VEIN ENDOTHELIAL CELL MITOGENESIS ASSAY

Human umbilical vein endothelial cells (HUVECs) in culture proliferate in response to VEGF treatment and can be used as an assay system to quantify the effects of KDR kinase inhibitors on VEGF stimulation. In the assay described, quiescent HUVEC monolayers are treated with vehicle or test compound 2 hours prior to addition of VEGF or basic fibroblast growth factor (bFGF). The mitogenic response to VEGF or bFGF is determined by measuring the incorporation of [^{3}H] thymidine into cellular DNA.

MATERIALS

HUVECs: HUVECs frozen as primary culture isolates are obtained from Clonetics Corp. Cells are maintained in Endothelial Growth Medium (EGM; Clonetics) and are used for mitogenic assays described in passages 1-5 below.

Culture Plates: NUNCLON 96-well polystyrene tissue culture plates (NUNC #167008).

Assay Medium: Dulbecco's modification of Eagle's medium containing 1 mg/mL glucose (low-glucose DMEM; Mediatech) plus 10% (v/v) fetal bovine serum (Clonetics).

Test Compounds: Working stocks of test compounds are diluted serially in 100% dimethylsulfoxide (DMSO) to 400-fold greater than their desired final concentrations. Final dilutions to 1X concentration are made directly into Assay Medium immediately prior to addition to cells.

10X Growth Factors: Solutions of human $VEGF_{165}$ (500 ng/mL; R&D Systems) and bFGF (10 ng/mL; R&D Systems) are prepared in Assay Medium.

10X [^{3}H]Thymidine: [Methyl-^{3}H]thymidine (20 Ci/mmol; Dupont-NEN) is diluted to 80 µCi/mL in low-glucose DMEM.

Cell Wash Medium: Hank's balanced salt solution (Mediatech) containing 1 mg/mL bovine serum albumin (Boehringer-Mannheim).

172°C. ^{1}H-NMR (500 MHz, $CDCl_3$): 8.26 (1H,s), 5.80 (1H,s), 5.24 (1H,m), 5.08 (2H, m), 3.76 (2H,m), 3.27 (2H,m), 1.96 (2H,m), 1.71 (2H, m), 1.48(9H, s). M+1 = 295.3

Synthesis of tert-butyl-4-({6-[(5-cyano-1,3-thiazol-2-yl)amino]pyrimidin-4-yl}oxy) piperidine-1-carboxylate **(1-2)**

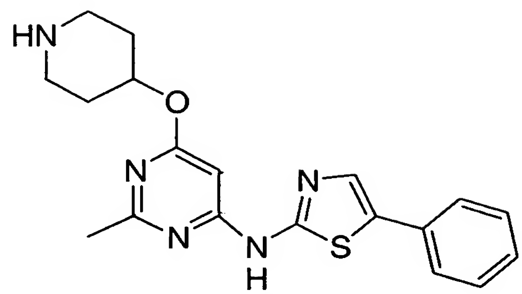
Sodium hydride (60% in mineral oil, 95 mg, 2.4 mmol) was added to a solution of tert-butyl-4-[(6-aminopyrimidin-4-yl)oxy]piperidine-1-carboxylate (295 mg, 1 mmol) in dry THF (3 mL). After gas evolution ceased (~1/4 hr) a solution of 2-chloro-1,3-thiazole-5-carbonitrile (162 mg, 1.1 mmol) dissolved in THF (2 mL) was added and the reaction was warmed to ~50°C for 22 hours. The cooled reaction was diluted with ethyl acetate and 2N HCl (0.7 mL) was added, followed by saturated $NaHCO_3$ solution. The ethyl acetate exact was dried over anhydrous Na_2SO_4, filtered and evaporated. The residue was chromatographed on silica gel eluting with a 0-2% methanol/chloroform gradient. The appropriate fractions were combined and the solvents removed. The resultant residue was triturated with diethyl ether to give the title compound as a white solid, mp: 224-227°C. ^{1}H-NMR (500 MHz, $CDCl_3$): 8.63(1H,s), 7.95(1H,s), 7.27(1H,s), 6.70(1H,br s), 5.36 (1H,m), 3.77(2H,m), 3.30 (2H,m), 2.05 (2H,m), 1.75 (2H,m), 1.48 (9H,s). M+1 = 403.2

Synthesis of 2-{[6-(piperidin-4-yloxy)pyrimidin-4-yl]amino}-1,3-thiazole-5-carbonitrile **(1-3)**

A solution of tert-butyl-4-({6-[(5-cyano-1,3-thiazol-2-yl)amino] pyrimidin-4-yl}oxy)piperidine-1-carboxylate (56 mg, 0.14 mmol) in methylene chloride (5 mL) containing TFA (0.5 mL) was stirred at room temperature over night. The solvents were evaporated and the residue was chased with chloroform (2X). The residue was dissolved in methanol, filtered through a plug of charcoal and the solvent nearly all removed under vacuum. This residue was triturated with ethyl acetate to give the title compound as a crystalline TFA salt, mp: 240-242°C. ^{1}H-NMR (500 MHz, DMSO-d_6): 8.68(1H,s), 8.33(1H,s), 6.41(1H,s), 5.33(1H,m), 3.26(2H,m), 3.13 (2H,m), 2.11(2H,m), 1.86(2H,m). M+1 = 303.2

Compounds **1-4** through **1-9** were synthesized via the same protocol shown in Scheme 1 by using an appropriate alcohol and 5-substituted 2-chlorothiazole.

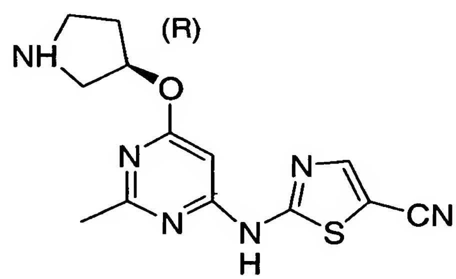
Synthesis of tert-butyl-4-({6-[(5-phenyl-1,3-thiazol-2-yl)amino]pyrimidin-4-yl}oxy) piperidine-1-carboxylate **(1-4)**



2-4

The title compound was prepared according to the procedure described in Scheme 2 substituting 2-chloro-5-phenyl-1,3-thiazole for 2-chloro-1,3-thiazole-5-carbonitrile. This product was obtained as the TFA salt, mp: 247-249°C. ^{1}H-NMR (500 MHz, DMSO-d_6): 8.52(1H,br s), 8.45(1H,br s), 7.82 (1H,s), 7.61 (2H, d, J=7.3Hz), 7.41(2H, t, J=7.6 Hz), 7.29 (1H, t, J=7.6Hz), 6.24 (1H, s), 5.31 (1H,m), 3.25 (2H, m), 3.15 (2H,m), 2.54(3H,s), 2.11 (2H,m), 1.87 (2H,m). M+1= 368.2

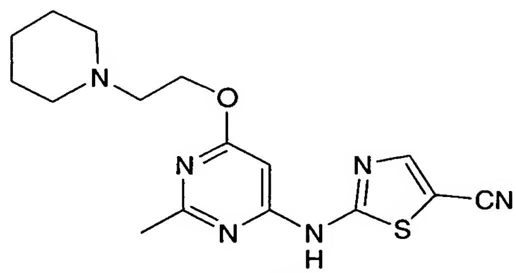
Synthesis of 2-({2-methyl-6-[(3R)-pyrrolidin-3-yloxy]pyrimidin-4-yl}amino)-1,3-thiazole-5-carbonitrile (2-5)



2-5

The title compound was prepared according to the procedure described in Scheme 2 except tert-butyl-(3R)-3-hydroxypyrrolidine-1-carboxylate was substituted for tert-butyl-4-hydroxy-piperidine-1-carboxylate. This product was obtained as the TFA salt, mp: 221-223°C. ^{1}H-NMR (500 MHz, DMSO-d_6): 9.05(1H,br s), 8.90(1H,br s), 8.32 (1H,s), 6.24 (1H, s), 5.67 (1H,m), 3.48 (1H, m), 3.41 (1H,m), 3.33(2H, m), 2.58(3H,s), 2.26 (1H,m), 2.17 (1H,m). M+1= 304.2

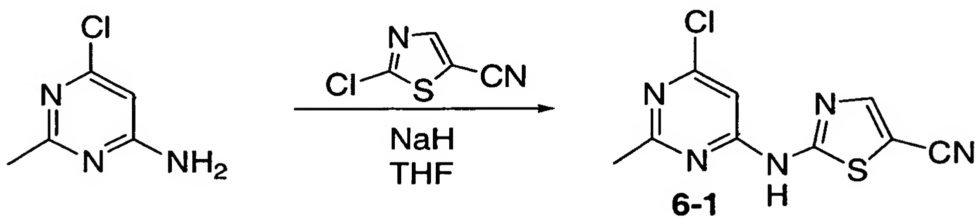
Synthesis of 2-({2-methyl-6-[(3S)-pyrrolidin-3-yloxy]pyrimidin-4-yl}amino)-1,3-thiazole-5-carbonitrile (2-6)



5-4

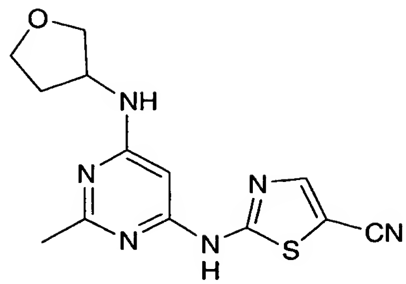
The title compound was prepared according to Scheme 5, except 2-piperidin-1-ylethanol was substituted for 3-morpholin-4-yl propan-1-ol to obtain 2-{[2-methyl-6-(2-piperidin-1-ylethoxy)pyrimidin-4-yl]amino}-1,3-thiazole-5-carbonitrile as the trifluoroacetic acid salt. ^{1}H-NMR (500 MHz, CD_3OD): 8.04 (1H, s) 6.25 (1H, s), 4.75 (2H, t, J=5.13hz), 3.66 (2H, d J=12.21Hz), 3.57 (2H, t, J=5.12Hz), 3.05 (2H, t, J=12.69Hz), 1.97 (2H, d, J=15.14Hz), 1.82 (3H, m), 1.54 (1H, m). M+1 = 345.0

SCHEME 6



<u>Synthesis of 2-[(6-chloro-2-methylpyrimidin-4-yl)amino]-1,3-thiazole-5-carbonitrile **(6-1)**</u>

6-Chloro-2-methylpyrimidin-4-amine (144.0 mg, 1.0 mmol), which is prepared from 6-amino-2-methylpyrimidin-4-ol as described in *Chem. Ber.*, 75, 755(1942), was added to the reaction flask along with dioxane (3.0 mL), and sodium hydride (60% in mineral oil, 120 mg, 3.0 mmol). After the effervescence had subsided 2-chloro-1,3-thiazole-5-carbonitrile (145.0 mg, 1.0 mmol) was added. After 2.0 hours the heat was removed and the reaction allowed to stir over night. The reaction was diluted with water then, acidified with concentrated HCl. The pH was adjusted to 8 with sodium bicarbonate and the aqueous layer extracted with ethyl acetate (3x100mL). The combined extracts were washed with brine (1x25mL) and dried ($MgSO_4$). Solvent removal yielded the desired 2-[(6-chloro-2-methyl-pyrimidin-4-yl)amino]-1,3-thiazole-5-carbonitrile. ^{1}H-NMR (500 MHz, DMSO-d_6): 12.74 (1H,s), 8.38 (1H,s), 6.95 (1H, s), 2.62 (3H, s). M + 1=252



8-8

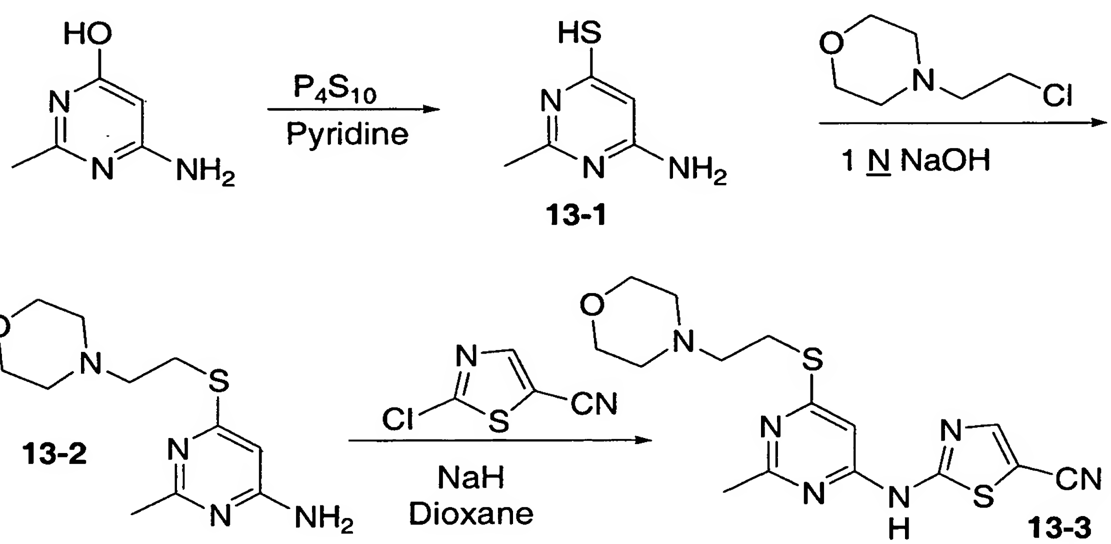
2-[(6-Methoxy-2-methylpyrimidin-4-yl)amino]-1,3-thiazole-5-carbonitrile (100.0 mg, 0.397 mmol) was added to a reaction flask along with tetrahydrofuran-3-amine hydrochloride salt (136.3 mg, 0.79 mmol), N-ethyl-N,N-diisopropylamine (410 μL, 2.38 mmol) and n-butanol (3.0 mL). The reaction was heated at reflux for one week. The reaction was cooled and the n-butanol was removed. The residue was treated with trifluoroacetic acid (0.5 mL) and diluted with methanol (3.0 mL). The mixture was then purified on a preparative high pressure chromatograph. This yielded 2-{[2-methyl-6-(tetrahydro-furan-3-ylamino)pyrimidin-4-yl]amino}-1,3-thiazole-5-carbonitrile as the trifluoroacetic acid salt. ^{1}H-NMR (500 MHz, DMSO-d_6): 11.99 (1H, br s), 8.23 (1H, s) 7.65 (1H, br s), 5.93 (1H, br s), 4.98 (1H, br s), 3.83 (2H, m) 3.71 (1H, m), 3.52 (1H, br s), 2.42 (3H, s), 2.16 (1H, m), 1.80 (1H, br s). M+1 = 303

NMR (500 MHz, CDCl3): 8.48(1H, s), 8.46 (1H, br s), 7.94 (1H, s), 5.36 (1H, m), 3.74 (2H, m), 3.36 (2H, m), 2.15 (3H, s), 2.00 (2H, m), 1.77 (2H, m), 1.48 (9H, s). M+1 = 417.3

Synthesis of 2-{[5-methyl-6-(piperidin-4-ylamino)pyrimidin-4-yl]oxy}-1,3-thiazole-5-carbonitrile (**12-2**)

A solution of tert-butyl-4-({6-[(5-cyano-1,3-thiazol-2-yl)amino]-5-methylpyrimidin-4-yl}oxy)piperidine-1-carboxylate (64 mg, 0.15 mmol) in chloro-form (6 mL) containing TFA (2.0 mL) was stirred at room temperature over night. The solvents were evaporated and the residue was chased with chloroform (2X). The residue was dissolved in a minimum amount of methanol and diluted with diethyl ether and the title compound crystallized out as the TFA salt, mp: 214-216°C. ^{1}H-NMR (500 MHz, DMSO-d$_6$):11.8 (1H, br s), 8.56(1H, s), 8.52(1H,br s), 8.36 (1H, s), 5.36(1H,m), 3.25(2H, m), 3.18 (2H,m), 2.19(3H, s), 2.12 (2H,m), 1.91 (2H,m). M+1 = 317.3

SCHEME 13



Synthesis of 6-amino-2-methylpyrimidine-4-thiol (**13-1**)

The title compound was prepared by modification of the procedure described Chem. Pharm. Bull (Tokyo), 11, 912-917 (1963) [Chem. Abstr. 60:1748a]. Phosphorus pentasulfide (12.2 gm, 50 mmol) was added to pyridine (80 mL) and stirred for 30 minutes. Then 6-chloro-2-methylpyrimidin-4-amine (2.5 gm, 20 mmol) was added and the mixture was heated at 110°C for 24 hours. The pyridine was removed under reduced pressure and the yellow solid suspended in water (50 mL). 5 N NaOH was added until nearly all solid dissolved in the